Chapter 17
Forensic Entomology

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Contents

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17.1 Introduction

Entomology in the strict sense is the study of insects. Insects are invertebrate animals in the Phylum *Arthropoda* and contained in the superclass *Hexapoda.* This superclass is variously divided by different authors but the most common division is into two classes: *Entognatha* and *Insecta*. The vast majority of the species are found in the class *Insecta*. As a group, the *Hexapoda*, commonly all referred to as "insects," are probably the single most successful and numerous animals on earth. To date, there have been approximately 900,000 species described, and it is estimated that this number reflects only a small portion of the actual species in existence. At present, there are over three times as many insect species described as for all other groups of animals combined. By way of example, in the Nearactic region alone, there are an estimated 125,000–150,000 known species of insects. By contrast, there are only approximately 3,200 species of mammals described worldwide.

Insects are found in virtually every habitat imaginable, with the exception of the depths of the oceans, and show a wide variety of adaptations to different habitats and food resources. While the vast majority of species of insects is not directly involved with human activities and are a significant part of the world's ecosystems, a number of species are in direct competition with man for food resources and cause millions of dollars loss to food crops on an annual basis. Still others are vectors of pathogens causing human disease or are themselves the etiologic agents. The diseases transmitted are among the most serious and include epidemic diseases such as Bubonic Plague and Yellow Fever. Additionally, some species of insects may cause harm to humans and animals through their bites and stings.

Insects are also beneficial. They serve as pollinators of plants, including those used by man for food, are parasites and predators of a number of species of organisms considered pests due to their activities, and are the major recyclers of dead plant and animal materials in the ecosystem. Additionally, they produce many products used by humans, including bees' wax, honey, and silk. Insects have been used extensively in scientific studies and have contributed to many advances in human and animal health. In many parts of the world, insects are used as food and are a significant source of protein. Without insects, the world as we know it would cease to exist.

Insects include well-known animals, including the cockroaches, flies, beetle, butterflies, grasshoppers, termites, ants, and bees. As adults, all have a body with an external skeleton (exoskeleton) that is divided into three major sections: head, thorax, and abdomen. The adults have three pairs of jointed legs that are located on the thorax and most species have one or two pairs of wings. The head has a pair of antennae and most species also have a pair of large compound eyes.

The development of an insect involves a series of different, distinct stages, termed metamorphosis. The majority of insects lay eggs, although some species, such as the Flesh Flies, produce live young. In some species, the eggs hatch into immature forms that are quite similar to the adults but lack wings. In these, the wings develop, showing as pads on the external surface of the body. This type of development is often termed an Incomplete Metamorphosis and several different types are recognized. In most insects, the eggs hatch into an

immature form that does not at all resemble the adult. These immatures, called larvae, pass through a series of stages, termed instars, until they have reached a maximum size. At that point, the larvae enter an inactive stage called a pupa, often passed within a protective casing. During the pupal stage, the insect essentially undergoes a second embryology and the winged adult insect emerges. This type of metamorphosis is termed Holometabolous or Complete Metamorphosis. The majority of insects involved in forensic entomology show Holometabolous Metamorphosis.

The earliest recorded case of the use of insects in legal investigations comes to us from thirteenth century China. In a book written by the Chinese death investigator, Sung Ts'u, titled "The Washing Away of Wrongs," a case is detailed of a murder in a village. The victim had been slashed to death and the wounds were reminiscent of sickle wounds. Because questioning of villagers and other investigative methods were not yielding any results, the magistrate had all of the villagers assemble, each bringing his own sickle. In the summer sun, flies congregated on one sickle that still had fragments of tissue and blood present. Confronted with this, the owner confessed to the crime. While this, at first glance, may seem relatively basic, it does indicate that the magistrate had considerable knowledge of the behavior and activity patterns of flies. Following this beginning, use of insects in criminal investigations was spasmodic at best, with records appearing of cases in Europe during the mid-1800s and later in the United States and England. Generally these were accounts of a specific case and, once the case was solved, the entomology faded into the background. It was not until the mid-1980s that forensic entomology began to develop as a distinct discipline in the United States, and a core of researchers began to meet routinely to discuss cases and exchange information. This eventually led to the formation of the American Board of Forensic Entomology and, later, the North American Association of Forensic Entomologists. An account of the recent development of forensic entomology in the United States is given by Goff [9].

In the broadest sense, forensic entomology is concerned with all cases in which insects become evidence in legal proceedings. From a practical standpoint, it is subdivided into three subdisciplines: stored product/urban entomology; structural entomology; and medicolegal or medicocriminal entomology. Of these, stored product/urban and structural entomology are typically involved in civil litigation, while medicolegal entomology involves situations in which a crime has been committed.

An example of a *stored product situation* would be a case of insect contamination of a box of breakfast cereal. The box is purchased at a local market and, when opened, it is found to contain insects along with the cereal, occasionally, more insects than cereal. The consumer, assuming they are not entomologists, does not like this and returns the box to the market for a refund. If enough boxes are returned, the market owner seeks to recoup his losses from his supplier. The entomologist is brought in to determine the type of insect involved, what was not done to prevent the infestation, and possibly assist in assigning blame. The bottom line in this situation is money and often involves significant amounts past simple replacement of the product. Typically, the entomologist is brought into the case relatively late in the investigation.

The situation in *structural entomology* is similar, but the problem involved is insect damage to human dwellings. An excellent example of this can be found in termite damage to houses. Typically, in areas known to have termite problems, preventive measures are taken during the construction of a house. These can take the form of physical or chemical barriers to prevent termites from invading and destroying the structure. Some of these are permanent barriers, while others, such as chemical ground treatments, may have a limited period of effectiveness and must be repeated on a routine basis. If the barriers fail and termites invade the structure, the entomologist is brought in to determine what went wrong and, again, assist in assigning blame. The ultimate solution again is financial.

In *medicolegal* or *medicocriminal entomology*, a crime has been committed and monetary considerations are generally absent. Unlike the previous subdivisions, the entomologist is typically contacted and involved in the investigation during the early stages, frequently working in cooperation with police and/or medical examiners. Insects in this context can be of assistance in several ways, including estimations of the period of time since death, determining postmortem movement of a body, assisting in assessment of wounds present on the body, characterization of the crime scene, as alternate specimens for toxicological analyses, as alternate sources of DNA, and in evaluation of cases of abuse and/or neglect of children and the elderly. Each of these will be discussed in this chapter.

In the vast majority of cases, the insect evidence only becomes of significance after a period of longer than 24 hours following death. In order to understand the applications of insect evidence, it is necessary to have some understanding of decomposition.

17.2 Decomposition

Most of the data used in the study of decomposition have been derived from controlled studies. The majority of these studies have been conducted as ecological studies and using test animals other than humans. The size of the animals involved has ranged from frogs and toads (Cornaby, 1974) to elephants [6]. Significant work using human cadavers has been conducted at the Anthropological Research Facility of the University of Tennessee at Knoxville. Among these are the early work by Rodriguez (1986) specifically looking at insect activity associated with decomposing bodies and later work by Schoenly et al. [23]. As early as 1989, the domestic pig was suggested as being a standard surrogate for humans in decomposition studies and this was reinforced by work by Schoenly et al. [23].

A common theme throughout the majority of these studies has been a division of decomposition into a series of discrete stages. While decomposition is, in reality, a continuous process, there is a value to establishe these, admittedly artificial, stages. Decomposition does not take place as a series of discrete combinations of physical parameters associated with distinct assemblages of arthropods. Use of stages does assist when faced with the problem of explaining a complex process to a jury.

While studies have been conducted in a variety of geographic locations and ecological situations, the major groups of arthropods involved remain similar from one area to the next. While the major groups remain constant, there are localized variations in the species within each group involved with the exception of some taxa having a wide geographic distribution. The number of stages proposed has ranged from two to as many as eleven (Goff, 1993). Regardless of location, a generalized pattern of five stages of decomposition has been established [8] that is applicable for most existing studies. These stages have seen a relatively wide acceptance, although the names of the stages have been modified by some workers [1, 24].

17.2.1 Fresh Stage

The fresh stage begins at the moment of death and is considered to end when the first signs of abdominal bloating are first observed. The first insects to arrive at the corpse following death will be adult flies. These are in the families *Calliphoridae* (Blow Flies and Bottle Flies) and *Sarcophagidae* (Flesh Flies). The female flies will typically go to the natural body openings. These will primarily be those on the head, with openings of the anus and genitals as secondary sites of attraction. If wounds are present on the body, inflicted prior to or at the time of death, these will also be attractive. Postmortem wounds are not as attractive to the flies. The female flies will enter the body openings and either lay eggs or deposit live larvae, depending on the species involved.

17.2.2 Bloated Stage

When the gasses produced by the metabolic activities of the anaerobic bacteria first begin to produce a slight inflation of the abdomen, the bloated stage is considered to begin. The continued actions of these bacteria will eventually result in the body becoming fully inflated and having a balloon-like appearance. During this stage, the internal temperatures of the body begin to rise as a result of the processes of putrefaction and the metabolic activities of maggots feeding inside the body. Species in the family *Calliphoridae* are strongly attracted to the body during this stage. As internal pressures increase as a result of the production of gasses, fluids are forced from the natural body openings and seep into the soil beneath the body. These fluids, in combination with the ammonia produced by maggot activities, result in the soil under the body becoming strongly alkaline and the normal soil fauna departs, to be replaced by a fauna specific to decomposition.

17.2.3 Decay Stage

The decay stage is the only stage of decomposition that has an actual physical event marking the start point. When the feeding activities of the maggots penetrate the outer surface of the body, allowing the gasses produced to escape, and the body deflates, the decay stage is considered to begin. The predominant feature of this stage is the presence of large feeding masses of *Diptera* larvae on the body. While predatory species, such as beetles in the families *Staphylinidae* and *Histeridae*, were present during the fresh and bloated stages, they show a marked increase in number during this stage. These, along with late-arriving necrophages, are observed in large numbers during the later portions of this stage. During the decay stage, *Diptera* larvae begin to complete their development and leave the body to pupariate in the soil adjacent to the body. By the end of this stage, most of these will have completed their development and departed. Most of the flesh will have been removed from the body by the end of this stage, and adults of taxa feeding primarily on dried skin and cartilage will arrive.

17.2.4 Postdecay Stage

As the body is reduced to dried skin and cartilage and *Diptera* larvae cease to be the predominant taxa present, the postdecay stage begins. There is no specific start point to this stage and the onset will be determined by local environmental conditions and judgment of the worker. Various species of *Coleoptera* will be the predominant feature of this stage in xerophytic and mesophytic habitats. This will increase in terms of both total numbers and species diversity as the stage progresses. In wet habitats, as encountered in swamps and tropical habitats, the body does not dry sufficiently for many of the *Coleoptera* species to exploit the body and these are functionally replaced by additional species from other groups, such as the *Diptera*. Associated with this in all habitat types is an increase in the number and diversity of the predators and parasites of the respective groups.

17.2.5 Skeletal Stage

The skeletal stage is reached when only hair and bones remain. There are generally no obvious carrion-frequenting taxa present and, as time passes, there will be a gradual return of the normal soil fauna and departure of the fauna associated with decomposition. During the early portions of this stage, there is a characteristic acarine fauna present in the soil under the body. While this fauna has great potential, effective use is currently hampered by the lack of adequate baseline data concerning many species and the great diversity of populations over even limited geographic areas (Schoenly & Reed, 1987). There is no definite end point to this stage. Localized variations in the fauna, particularly the soil fauna, may be detectable for months or even years following death (Goff, 1989, 1991).

17.3 Basis for Use of Insects

Insect invasion of an exposed body can begin shortly following death, often in as little as ten minutes for an exposed body under summer conditions. In the majority of cases, insect evidence is employed after a period of 24 hours. Prior to this, other techniques are available for assessment of the body. In order to understand the use of insects, it is necessary to understand the relationships of insects to a decomposing body. A dead body is a temporary disruption of the ecosystem and presents a progressively changing food resource to a variety of organisms, including insects, algae, fungi, and a variety of microorganisms. Insects encountered on a decomposing body will consist of elements of the insect fauna unique to the habitat in which the body is found as well as insect species specific to the decomposition process. Both types will have an application in investigations. There are five types of relationships of insects to the decomposing body.

17.3.1 Necrophagous Species

Necrophagous species are insects and other arthropods that feed directly on the decomposing body. These include many of the *Diptera* or true flies, as well as many beetles. Species found in this group are often the most significant taxa used in the estimation of the period of time since death or PMI during the early stages of decomposition, days one to fourteen. It must be kept in mind that entomological estimates of the PMI are actually estimates of the minimum period of insect activity on the body and not the actual interval itself.

17.3.2 Parasites and Predators

This is generally accepted as the second most significant group of carrion-frequenting taxa (Lord & Goff, 2003). As the initial invading insects feed on the body and thus alter the body, they make it attractive to other groups of arthropods. Among these are those insects that are predatory or parasitic on the necrophagous insects. In some instances, as is the case for some *Diptera* species, larvae that are necrophagous during the early stages of development become predators during the later stages. An excellent example of this is found in the Hairy Maggot, *Chrysomya rufifacies*. Taxa included in this category are *Coleoptera* (beetles), *Diptera* (true flies) and *Hymenoptera* (bees and wasps) parasitic on larvae and pupae.

17.3.3 Omnivorous Species

Included in the omnivorous species category are insects that feed both on the body and associated arthropods. These include species of ants, wasps, and some beetles. It should be noted that large populations of these, particularly ants, may

actually exert enough pressure on the necrophagous species to retard the rate of carcass removal [8].

17.3.4 Adventive Species

In the adventive species category, we place those species that use the decomposing body as an extension of their natural habitat. This includes species of *Collembola*, spiders, and centipedes. There is no actual relationship to the body except that it can serve as a concentrating mechanism for those animals or plants that are the normal food source for these species. During the later stages of decomposition, these species tend to concentrate in the substrate under the body. Mites in the families *Acarideae*, *Lardoglyphidae*, and *Winterschmidtiidae*, feeding on molds and fungi growing on the body, are included in this category. There are also a large number of mites in the suborder *Gamasida* associated with a decomposing body, but their relationships to the body are not clearly defined (Goff, 1989). These include species in the families *Macrochelidae*, *Parasitidae*, *Parholaspidae*, *Cheyletidae*, and *Raphignathidae.*

17.3.5 Accidental Species

One group often overlooked or assigned disproportionate significance is the accidental species. These insects do not have any relationship to the body but just happen to be there. It seems strange to some workers but, when an insect stops flying, it has to sit on something. Sometimes that is a body. We must also include in this class those insects normally associated with plants that may be in close proximity to a body. When vegetation is disturbed, many insects respond by dropping to the ground. In this type of situation, as the body is moved, insects may fall onto the body from surrounding vegetation and later be found on the body. Common sense and some knowledge of the insects should allow for resolution of these situations.

17.4 Collection of Entomological Evidence

While the insects and other arthropods associated with a dead body are potentially a valuable tool, their ultimate utility depends on proper collection and preservation for later analyses. The techniques involved are not particularly complicated and can be done with a minimal effort if some preparation is done beforehand. Obviously, the ideal situation is to have a trained entomologist examine the body and make the collections. This is generally not possible, so simple but effective techniques are presented here to assist the non-entomologist.

17.4.1 Equipment Needed

Surgical gloves
Forceps
Glass vials
Trowel
Insect net
Artist's brush
Paper bags
Ice cream cartons or their equivalent
KAA or other insect fixative
Vermiculite or sand
Ethyl alcohol (EtOH) (70–80%) or isopropyl alcohol
Can of cat or dog food

17.4.2 Collection Procedures

The insects collected will be divided into several groups, depending on where they are found on the body: flying, crawling, and soil-dwelling. Before beginning any collections, it is advisable to have people move away from the body for approximately ten to fifteen minutes. When a body is disturbed, as in processing, insects will leave the body. Because the insects of interest are largely dependent on the body as a food resource, typically, they do not move far from the body and will return relatively quickly once the activity ceases. It is important to collect flying insects as soon as possible. These are more mobile and will leave the body permanently if disturbed for a period of time. Collection is best accomplished using the insect net. While the net is relatively easy to use, I suggest that one should practice before attempting to collect at a scene. After all, you have just produced an insect net and you need to preserve some shreds of your dignity. Collections should be made by sweeping the net over the body without hitting the body. Collections should also be made from any bushes or vegetation surrounding the body. Many insects will move to vegetation adjacent to the body when initially disturbed. The insects collected in the net will typically be adults and hard bodies. These should be preserved in the EtOH (70–80%) in the glass vials. If isopropyl alcohol is used, it is generally 70% from the container and should be diluted 1:1 with water. Otherwise the specimens will become hardened and difficult to examine. Samples taken from different areas of the body and different vegetation should be kept separate and labeled as to origin, collector, manner of collection (net, etc.), and the date and time of collection.

Crawling insects are found on, in, and around the body. Collections must be as compete as possible to ensure that a representative sample of everything in the body is obtained. It is also important to remember to keep insects from different parts of the body separate and label as to their origin. In addition to the fact that the distributions of the insects on the body may prove to be significant to the entomologist in

arriving at the estimates, many species will feed on each other and other species of insects on a body. Crawling insects can easily be collected using forceps or by hand. Always wear surgical gloves while collecting. Aside from the obvious need to prevent contamination, some of the insects present may bite or sting. The arthropods collected will be either hard-bodied adults or relatively soft-bodied immature forms. While the hard-bodied insects can be treated in the same manner as the flying insects, the immature forms, or larvae, require special treatment to ensure that they will be in a condition suitable for analysis by the entomologist.

Immature insects are often difficult to identify to the species level and this is necessary in order to arrive at an accurate estimate. Even closely related species may have quite different rates and patterns of development. By contrast, most of the adults are easily identifiable by a trained entomologist. For this reason, it is a good idea to keep a portion of the immatures collected alive to be reared to the adult stage. Generally, the specimens are split into two parts. One is to be killed and preserved; this stops the biological clock. The second part is kept alive and allowed to reach the adult stage for easier confirmation of species.

The outer layer of the insect's body is a wax layer that assists in maintaining water balance. In order for proper preservation, this layer must be broken down to allow the preservative to enter the body. A number of different solutions that will serve this purpose are available. The most common is KAA. A KAA solution consists of 1 part glacial acetic acid, 1 part refined kerosene, and 30 parts 95% ethyl alcohol. Immature insects should be killed and placed into this solution for a period of five to ten minutes and then removed and stored in 70% EtOH or isopropyl alcohol diluted 1:1 with water. If the insect is allowed to remain in the KAA solution, it will eventually expand to the point of bursting and be of no use. If KAA is not available, an easy substitute treatment is to put the insect into hot water (76.7°C or 170°F) for a period of two to three minutes and then transfer the insect into 70% EtOH. Water close to this temperature can be obtained at most fast food restaurants by ordering either tea or decaffeinated coffee.

Rearing of immature insects to the adult stage requires an adequate food source and a set of controlled conditions. For this reason, the specimens collected at the scene should be transported to the entomologist as quickly as possible. The specimens collected should be placed into ventilated containers, most often cardboard ice cream cartons or their equivalent. The best size seems to be a half-pint container filled to approximately one quarter to one half with vermiculite or some other inert substance. It is advisable to add food to this container, and canned dog or cat food serves this purpose well. These containers and/or live specimens should not be placed into plastic bags under any circumstances. If the specimens cannot be shipped immediately to an entomologist, the insects can be reared in the cardboard containers. In this case, the food source should be placed into a watch glass and this placed inside the container. The top of the container should be covered with a fine gauze or organdy material, held in place by a rubber band. The food supply should be checked daily and additional food added as needed. The container should not be unduly disturbed. The larvae will complete their development inside the watch glass and then migrate away from the food source

and burrow into the vermiculite to enter the pupal stage. From this stage, the adults will emerge.

Once they have emerged, the adult insects should be supplied with food and water. The simplest way to accomplish this is to place a cotton ball soaked with water and sugar into the container. Adults should be killed after a period of 24 hours and preserved in 70% EtOH or pinned and dried. During the rearing process, the temperatures should be as close to those encountered at the scene as possible. Records of daily maximum/minimum temperatures should be kept and supplied to the entomologist.

It is essential that each lot of insect specimens be correctly and completely labeled to ensure that the evidence can be properly interpreted by the entomologist. Toward this end, there must be a label for each separate lot of specimens. The label should include the following information:

Date and time collected
Location of the body, as specifically noted as possible, geographic location, and type of terrain
Type of habitat in which the body is found – indoors, outdoors, vegetation type, dump, etc.
Location of the specimens on the body
Name, address, and telephone number of the collector

There should also be information concerning the body, including:

Sex, height, weight
Presence or absence of clothing; description of clothing
Orientation of the body – sitting, lying down, on side, back
Attempts to conceal body – wrapping, burial, in container, etc.
Physical damage to body
Cause of death if known
State of decomposition
Description of insect fauna associated with the body

Anything unusual about the scene should also be noted and a complete photographic record made of the scene. As with all other types of evidence, a proper chain of custody must also be maintained for the insect specimens.

17.5 Applications of Entomological Evidence

As noted earlier, there are a number of different situations in which insect evidence can be employed in criminal investigations. Insect evidence is obviously not present in all crimes and, even when present, may ultimately prove to be of minor significance to the resolution of the case. When the evidence is present, it does have the potential to be a powerful tool in many instances. Each of the known possible applications will be covered here.

17.5.1 Estimation of the Postmortem Interval

In practice, the major use of entomological evidence is in the estimation of the period of time since death or the postmortem interval, often abbreviated as PMI. In consideration of this, two major ideas must be kept in mind. First, this is an estimate not a precise time or date. Any estimate that does not include a range of possible times must be viewed with great suspicion. Second, what the entomologist actually calculates is an estimate of the period of insect activity on the body. This is not a calculation of the actual time of death, although, in the majority of cases, the two may be quite close. Generally speaking, the estimates are presented in terms of a minimum period of activity and the parameters of the estimates are directly proportional to the period of time since death. There are two basic approaches to the estimation of the period of time since death: calculation based on development of individual species and use of succession studies.

For approximately the first fourteen following death, estimates are most frequently based on the development of species of *Diptera*. The adult flies are typically the first insects to arrive at a body, often as soon as ten minutes following death. The female fly is typically attracted to the natural body openings of the head, anus, and genitals. The female fly investigates these areas and, if the substrate is suitable, will deposit either eggs or live larvae. This starts a biological clock that is stopped when the body is discovered. During its development, a *Diptera* species passes through a distinctive series of stages leading to the adult stage (Fig. 17.1).

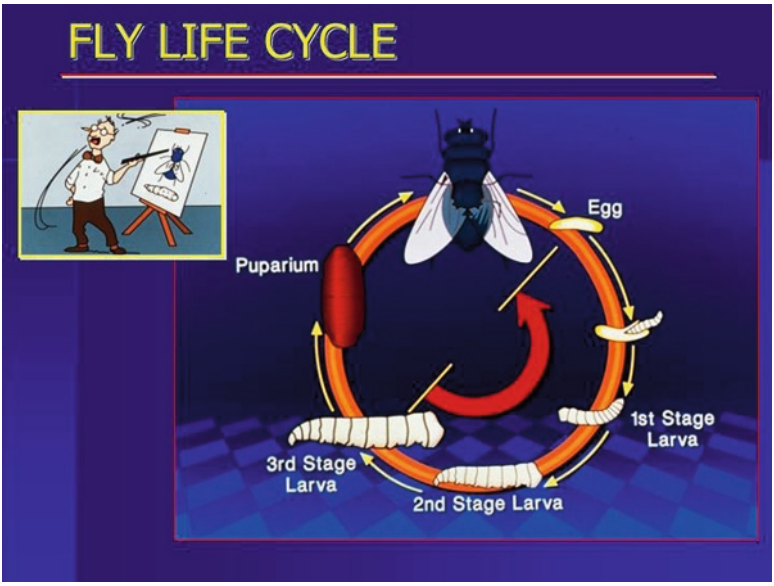

Fig. 17.1 Life cycle of a blowfly, Family *Calliphoridae*

When collecting, fixing, and preserving specimens from the body, the entomologist looks for the most mature specimens present on the body, but also collects a representative sample of all species and stages of development present. By determining the most mature specimens present on the body, the period of time required to reach that stage of development can be estimated by reference to previously conducted laboratory studies. This requires an accurate identification of both the species and stage of development.

One of the major problems facing an entomologist is the accurate identification of the larvae or maggots present. Often the entomologist will not be present at the scene and must rely on dead specimens collected by others. All too often, these arrive in marginal states of preservation and are not useful for identifications. The Appendix gives a protocol for collection of specimens that, if followed, will serve to ensure their arrival in a usable state, and instructions that are more detailed are given in Catts and Haskell [5]. Even when the local fauna is well known, there are significant problems in making accurate identification from only immature specimens, particularly during the early stages of development. Recent publications have provided keys for the identifications of larvae of forensically significant species [15, 22]. While these keys are presented as being somewhat regional in scope, given the relatively wide distribution of many forensically significant species, their applications are wider than indicated. Even with these keys, specimens must often be reared to the adult stage for accurate species-level identifications. Recent investigations of DNA analyses of larvae show considerable promise in identifications of immature specimens [25, 26].

In the calculation of the developmental times, considerable emphasis is placed on the relationships between ambient temperature and the durations of the different stadia. The rate of development and activity of insects, as is the case with other ectotherms, is directly related to temperature. As temperature increases, the rate of development increases and the durations of the individual stages in the life cycle become shorter. For example, the blow fly *Calliphora vicina* requires 11 days to complete the 3rd instar at 12.5°C but only 6.3 days at 22°C [15]. Additionally, there are also lower and upper temperature limits for development and activity of species. These temperature limits vary with the individual species and appear to also be subject to geographic variations within a given species. In the past, many workers have used a lower limit of 10°C as a lower limit for development. Unfortunately, the situation is more complicated, and the easy cutoff points used in the past do not appear to be as relevant as once thought. Fortunately, a number of different controlled studies on developmental rates have been conducted at different temperatures, so baseline data are available for most widely distributed species.

In estimating the PMI, it is necessary to convert time and temperature into units that can be used to compare laboratory data with conditions at a scene. The most common approach is to use the ecological concept of Degree Hours (DH) or Degree Days (DD). This technique was first developed to assist in predicting pest outbreaks in crops, but it has been adapted for use in forensic entomology. In this approach, ambient temperatures from weather stations in the vicinity of the body are used as an indication of the temperatures at which the larvae developed. In reality, this is an oversimplification of a very complicated situation, but, none-the-less, it is functional.

One problem lies in that, as the maggots feed and develop, they form feeding masses. Maggots found infesting a body have typically come from eggs that were laid at about the same time, hatched at about the same time, and then form feeding masses. Maggots are more efficient feeding as a mass, as opposed to feeding alone. These masses generate heat, and temperatures as high as 53°C have been recorded inside these maggot masses. At these temperatures, most species of maggots cannot survive for extended periods and they must rotate through the mass, feeding for a period of time in the center of the mass and then moving to the outside of the mass to cool, then returning to the center to feed. While the ambient temperature does not reflect the actual temperatures at which the maggots are feeding, it does exert an overall influence on the mass and thus allows the DH or DD concept to work for our purposes.

Another common problem lies in the proximity of the weather station used to the actual scene containing the body. Even short distances can result in significant differences in weather conditions. For example, a site near the top of a hill or on a north-facing slope may have significantly different temperatures from a weather station near the base of the hill due to exposure to sunlight and/or differences in elevation. Because relatively few bodies are found close to weather stations, there must be some mechanism to reconcile data from the station to the conditions at the crime scene. The most commonly employed method used is linear regression. In this process, weather data are obtained from a recording device placed at the scene of the discovery of the body for a period of at least five days. Temperature data from that station is compared with corresponding data from the established station and a regression equation developed to allow for adjustment of data. Keep in mind that the resulting temperatures are not actual temperatures, but rather the best approximation of what probably took place at the site.

In arriving at the estimated period of time since death, time in hours or days is converted into thermal units by multiplying time by the temperature in degrees Celsius, expressed as:

$$\text{ADH}\left(\text{accumulated degree hours}\right) = \text{Time}\left(\text{hours}\right) \times \text{Temperature}\left(^{\circ}\text{C}\right).$$

Thus for a period of 24 hours at 15°C, the total number of ADH value would be:

$$24\ \text{hours} \times 15^{\circ}\text{C} = 360\ \text{ADH}$$

This would be the simplest approach; however, there are temperatures below which insect development ceases. As noted above, these temperatures termed “base temperatures” will vary from species to species. To adjust for these, a base temperature is subtracted from the ambient temperature in calculations and the equation is modified as follows, using a cutoff point of 10°C as an example:

$$\text{ADH}_{(\text{base } 10)} = \text{Time}\left(\text{hours}\right) \times \text{Temperature } ^{\circ}\text{C} - 10^{\circ}\text{C}.$$

Thus the same conditions as previously used result in an $\text{ADH}_{(\text{base } 10)}$ value as follows:

$$24 \text{ hours} \times (15°\text{C} - 10°\text{C}) = 120 \text{ ADH}_{(\text{base } 10)}.$$

It has been found that different species of insects have different base temperatures and these may also vary throughout the geographic range. As a general practice, it is preferable to use laboratory studies conducted on populations collected in close geographic proximity to the crime scene. For example, there may be some variations between populations of *C. vicina* in the Northwestern United States and populations from Europe.

In calculating the estimated period of insect activity on the body, ADH values are summed, beginning with the point at which the collections were made and working backward until the required ADH value for the most mature specimens collected is reached. This will estimate the minimum period of insect activity on the body and frequently the PMI. For example, in a case, the most mature specimens collected from a body were determined to be 3rd instar larvae of the Hairy Blowfly, *C. rufifacies*, measuring 13–15 mm total length, mean = 13.7 mm. Based on laboratory rearing data at 26°C, it would require approximately 136 h to reach that stage of development or 3,588 ADH.

Table 17.1 gives the ADH values calculated from hourly temperature data obtained from a National Oceanic and Atmospheric Administration (NOAA) weather station at an airport approximately 0.5 miles from the scene. Because the values are summed for each day, the required value of 3,588 ADH is attained at 2200 on 9 April, giving an estimated minimum period of time for insect activity. Because in this case the body was lying exposed on the ground, this estimate would most probably closely approximate the period of time since death.

Table 17.1 Accumulated degree hours (ADH) values from weather data collected from National Oceanic and Atmospheric Administration (NOAA) weather station for period 15 to 9 April

Date		DH	ADH
April 15		494.6[a]	494.6
April 14		604.2	1,098.8
April 13		597.1	1,695.9
April 12		613.0	2,308.9
April 11		613.3	2,922.2
April 10		607.9	3,530.1
April 09			
Hour	Temp (°C)	DH	ADH
2,400	23.9	23.9	3,554.0
2,300	24.4	24.4	3,578.4
2,200	24.4	24.4	3,602.8[b]
2,100	25.0	25.0	3,627.8

[a] Specimens were collected and preserved at 2000 on April 15
[b] Value of 3,588 ADH required for development of *Chrysomya rufufacies* attained

After the first fourteen days following death, most of the initial colonizing *Diptera* species will have completed their development and departed the body to complete their development through the puparial stage into an adult fly. At this point, the estimation of the period of insect activity becomes more difficult. As previously discussed, exploitation of a body by insects and other arthropods follows a pattern of succession. As one group of insects uses the body as a food resource, their activities change the character of the body, making it attractive to another group of species. As this group feeds, they, in turn, alter the body, attracting another group of insects. These species are not all feeding directly on the body, and the various relationships between arthropods and a decomposing body were discussed earlier. At this point, it becomes necessary to make a complete collection and identification of all of the species present on the body. By comparing the results of these collections with data from controlled decomposition studies conducted in similar geographic and ecologically similar habitats, it becomes possible to arrive at periods of time during which insect colonization most probably occurred.

This case gives an example of how this technique can be applied. The largely skeletal remains of a white woman were discovered in a sugar cane field on the island of Kauai. The cause of death was determined to be homicide due to multiple stab wounds that had resulted in cut marks to the ribs. Time since death was an obvious issue. Although there was little flesh remaining on the body, there were a number of insects present. There were empty puparial cases of the *Calliphoridae C. rufifacies* on and near the body. This species is an early colonizer of human remains in Hawaii, often arriving within ten minutes following death. Given prevailing temperatures in the area, development from egg to emergent adult requires approximately eleven days. Oviposition takes place for the first 5 days following death. Because only empty puparia were present, this indicated a minimum period of sixteen days. Also present on the body were larvae of the *Piophilidae Piophila casei* (Fig. 17.2).

This species, also known as a Cheese Skipper, arrives at remains in Hawaii on day fifteen and completes its development to the puparial stage by day 36. The specimens collected were beginning to depart the body for puparialtion, thus

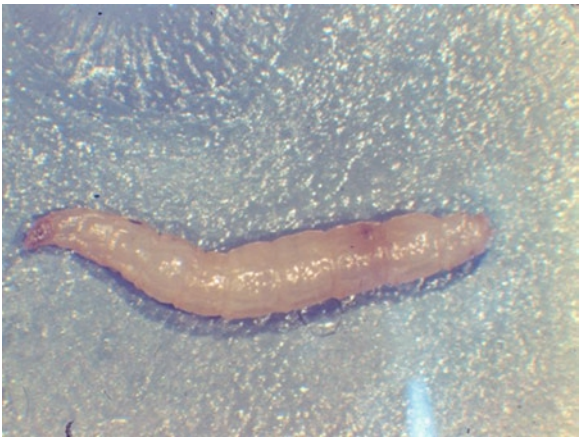

Fig. 17.2 Larva of the Cheese Skipper, *Piophila casei* (Family *Piophilidae*)

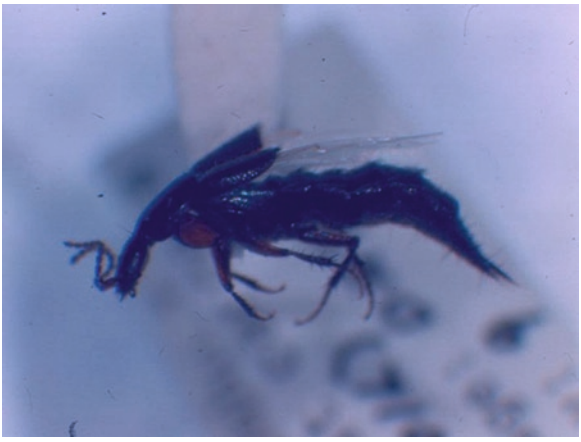

Fig. 17.3 Adult of the staphylinid beetle, *Philonthus longicornis* (Family *Staphylinidae*)

Fig. 17.4 Larva of the Black Soldier Fly, *Hermetia illucens* (Family *Stratiomyidae*)

indicating a time period of approximately 36 days. There were also *Staphylinid* beetles, *Philonthus longicornis*, adults present (Fig. 17.3).

These beetles in Hawaii arrive at the body approximately 25 days after death and leave by day 53. The last species identified was the Black Soldier Fly *Hermetia illucens* (Fig. 17.4).

This species is typically a late arrival, twenty days following death, and has a relatively long developmental period. The oldest specimens recovered were determined to be approximately fourteen days old, thus indicating a period of 34 days following death (20 + 14 days of development). Examining data for the species involved, an estimated period of 34–36 days following death was estimated. Although not a precise time, it was sufficient to allow for identification of a suspect who was eventually convicted of murder. While none of the species individually allowed for an accurate estimate, the combination of life cycles and behaviors of all collected proved significant.

As decomposition progresses and the analysis of the insect fauna moves from life cycles of a single species to patterns of succession, the parameters of the estimate become wider. For the first periods, the estimate is presented in terms of a range of hours, later changing to days, then months, seasons, and years. Finally, the only statement possible from an entomological standpoint is "the body has been there a long time." Regardless of the parameters, the estimate remains only that – an estimate – and an estimate of the period of insect activity on the remains, not the actual time since death. In many instances, this period is quite similar to the actual time since death. Regardless of the inherent limitations of the technique, it remains a powerful tool in the investigation.

17.5.2 Postmortem Movement of the Body

As noted previously, insects are among the most widely distributed organisms on earth and are the most numerous animals in the world. However, while insects are found virtually everywhere, individual groups and species of insects frequently have quite distinct patterns of distribution with respect to geography and ecology. In various decomposition studies conducted in a variety of different habitats and regions [1] (Carvalho et al., 2004; Goff, 1993), it has been observed that the general groups occur on a body in a similar pattern regardless of geography, the species composition will vary with the region. While many of the forensically significant species tend to have wider distributions, for example *C. vicina*, many species have narrow geographic ranges. Thus on any body, there will be species having wide distributions combined with species restricted to the particular habitat in which the body is found. Thus, if a body is discovered with insects restricted to a habitat or geographic region different from that in which it is discovered, this is an indication that the body may have been moved following death.

In addition to geographic differences, there may be habitat differences over a relatively short geographic distance. In one case in Hawaii [11], a body was discovered in a cane field in the middle of the island of Oahu. On the body there were three species of *Diptera* larvae. Two of these were in the family *Calliphoridae* and typical early colonizers of remains, *C. megacephala* and *C. rufifacies*. These were determined to be approximately 3.5 days into their larval development. The third species was in the family *Muscidae*, *Synthesiomyia nudiseta*, and showed a 5.5-day period of development. In Hawaii, both of the *Calliphoridae* species prefer an outdoor habitat, whereas the *Muscidae* is typically associated with indoor situations and is only rarely encountered outside of an urban situation. This two-day difference in development was an indication that the individual had been killed in an urban situation, probably indoors, the body was allowed to remain there for a period of approximately two days, and was then transported to the rural sugar cane habitat in which it was discovered. Subsequent investigation confirmed this scenario to be correct.

It is also possible to have insects not actually associated with the decomposition processes provide significant information. Many plant-feeding species have very

narrow food preferences and well as limited geographic ranges. If a body is outdoors near or under vegetation, it is possible for insects associated with that vegetation to move onto the body, although typically not to feed or lay eggs. For many foliage-feeding species, their defense relation to being disturbed is to release their hold on the plant and drop to the ground. If the body is between the insect and the ground, it may be moved with the body to the next location. Obviously this is not the normal occurrence, but it has been observed.

17.5.3 Assessment of Wounds

Insects colonize a decomposing body in a predictable manner [19]. The initial invasion is centered around the natural body openings. The primary site will be the openings of the head (nose, mouth, eyes, and ears), followed by the anus and genitalia, if exposed. This pattern is associated with the feeding behavior of the initial colonizing taxa, primarily *Diptera*. The mouthparts of their larvae or maggots are not constructed in such a manner as to be effective in penetration of intact skin, but are well suited to the mucus membranes associated with the natural body openings. Typically the third choice for colonization will be wounds inflicted antemortem or perimortem, while blood is still flowing. Wounds inflicted postmortem are not attractive to the *Diptera* for oviposition. As the maggots feed on the tissues associated with these antemortem or perimortem wounds, they change the surrounding tissues and a pattern of succession begins, similar to that observed with normal invasion of natural body openings. By contrast, activity associated with postmortem wounds is minimal by comparison. Because feeding activity by the insects often changes the appearance of the wounds, they may not be obvious as decomposition continues. By observing abnormal patterns of inset activity on the body, clues may be detected leading to discovery of otherwise obliterated wounds. For example, in one case, the body of a woman was discovered in the early portions of the decay stage of decomposition. Maggot activity was apparent in the area of her head. Additionally, there were centers of maggot activity observed in the chest and also the palms of both hands. As the skin, particularly in the palms of the hands, was normally not suitable for colonization, the pathologist investigated more closely and found evidence of stab wounds in the chest that had cut into the ribs. Wounds detected in the palms of the hand represented defense wounds. Thus, any deviation from the normal pattern of decomposition should be investigated, because significant information may be otherwise overlooked.

17.5.4 Crime Scene/Habitat Characterization

As noted previously in the discussion of postmortem movement, insects are often restricted in their geographic and seasonal distribution. These patterns can be of assistance in tying a suspect to a victim and/or crime scene. Webb et al. [21]

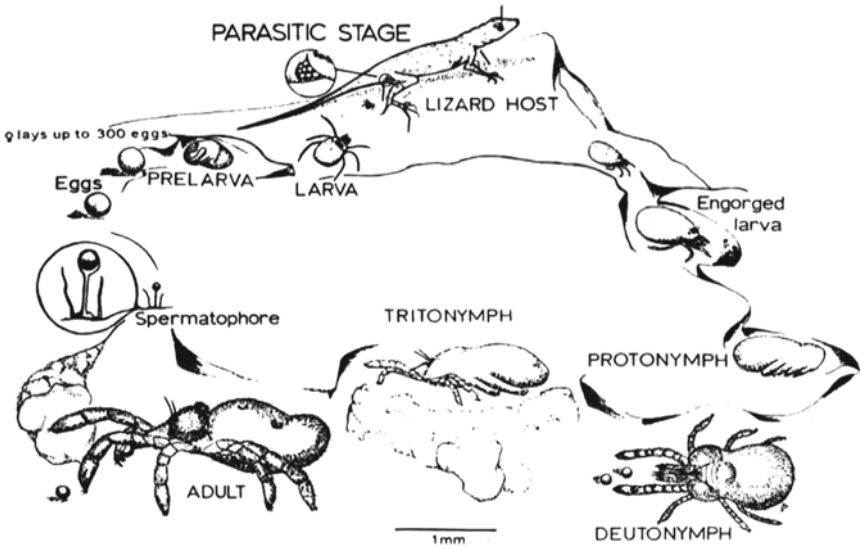

Fig. 17.5 Life cycle of a chigger mite (Family *Trombiculdiae*). Reproduced by permission of Entomological Society of America, Lanham, MD, © 1982

detailed a case from California in which a suspect was tied to a crime scene and victim based on the bites of a larval *trombiculid* mite or chigger. In this case the body was discovered in a scrub vegetation area in California. During the recovery process, most of the involved personnel were bitten by a pest species, *Eutrombicula belkini*. There were several suspects in the case, one with bites similar in shape and distribution on the body that would be anticipated from an attack by chiggers. The life cycle of a *trombiculid* mite is somewhat complex, with a combination of active and inactive stages, a parasitic habit, and a predatory habit (Fig. 17.5).

The larval stage is a parasite on vertebrates and is actually the only stage in the cycle properly termed a "chigger." Other stages are either predatory or inactive. The species tend to be quite habitat specific although exhibiting a wide host range, particularly the pest species biting man. In California, the distribution of chigger that will bite man is quite limited and the presence of the species at the recovery scene and bites on the individual served to tie him to the crime scene and thus the victim.

Another somewhat similar case was detailed by Greenberg (1993). In this case, an individual was tied to a rape by cockleburs found adhering to a ski mask. Access to the victim's bedroom was accomplished by the rapist climbing a tree and entering though a window. The suspect claimed that the ski mask had not been taken out of the drawer since the previous winter. The cockleburs on the mask were consistent with those found in the victim's yard, but the season remained in question. Closer examination of the seeds revealed the presence of beetle larvae that would be inconsistent with the mask having been worn only during winter months when the larvae would not be present. They were, however, consistent with the time of year of the rape.

17.5.5 Alternate Specimens for Toxicology

Often a body is discovered in an advanced stage of decomposition where tissues and fluids that would normally be sampled for toxicology are no longer suitable. In these situations, insects may still be present that can serve as adequate alternate samples for analyses. This subdiscipline of forensic entomology has been termed Entomotoxicology and is itself subdivided. One aspect deals with the simple detection of substances through analyses of insects associated with the body. The second area is more complicated and concerns itself with the effects of the substances in the decomposing tissues on the developmental patterns and rates of development of the insects using those tissues as a food source.

Among the first studies with this topic was that by Nuorteva and Nuorteva [17] dealing with environmental contamination by mercury as a means of determining geographic origin of a woman whose body was discovered in Finland. Subsequently, he demonstrated bioaccumulation of mercury in beetles feeding on maggots fed on tissues with a high concentration of mercury and developmental abnormalities associated with this bioaccumulation. Subsequent to this, beginning with Beyer et al. [2] a large number of toxins, pharmaceuticals, and illegal substances have been detected through analyses of maggots in both cases and experimental studies. Goff and Lord [14] summarized the work to date and subsequently Goff et al. [12],and Bourel et al. [3, 4] have added additional substances to the list. It must be noted that, although the various drugs, toxins and their metabolites have been successfully detected in larvae, there is no reliable technique allowing for determination of the actually dosage of the substance administered to the deceased. There are a number of different factors involved in this, including postmortem relocation of the drugs in the tissues, breakdown over time of the drugs and their metabolites, and depredation on the body by organisms other than insects. Although there have been some attempts at a correlation between concentration in tissues and concentrations in maggots, the results have not proven useful [16].

Maggot activity consists primarily of feeding and growing as rapidly as possible, with reproduction and dispersal left to the adult stage. This leads to the obvious question of "Is there an effect on the growth rate and/or pattern when the maggot ingests the drug or toxin along with the decomposing tissues?" In 1989, a body was recovered on the island of Oahu. Examination of the scene revealed evidence that the individual had been sitting on the limb of a tree overhanging a ravine and appeared to have died as the result of a fall. An estimated minimum period of insect activity was calculated. As the investigation continued, an individual came forward and stated she had seen and had a conversation with the deceased several days after the calculated minimum period. Because the deceased was a known cocaine abuser, the question arose as to the effect of cocaine in his tissues on the rate of development of maggots feeding on the body. As a result of this question, a controlled study was conducted and it was determined that maggots feeding on tissues from an experimental animal given a lethal dosage by

weight of cocaine did exhibit a significant acceleration in rate of growth [13]. Lower dosages did not have this effect. In the case in question, the dosage was not at a lethal dosage. The question of the conversation was resolved when it was discovered that the individual claiming to have had contact with the deceased had a history of similar claims and was not reliable. The original estimate of a minimum period was accepted.

17.5.6 DNA Applications

At present, applications of various techniques involving DNA analyses to forensic entomology are limited but the potential for significant uses in the future exist. The potential applications fall into two main areas: identification of species during early instars, and linking suspects to victims based on DNA present in gut contents.

It is a simple fact that, with relatively few exceptions, there is a marked similarity between maggots during the 1st instar, even to a trained entomologist. As noted earlier, there are also significant differences in the durations of the different stages of the life cycle even among closely related species. For this reason, accurate identification of the species becomes essential for an accurate estimation of the period of activity on the body. At present, the most commonly employed technique is to rear a portion of the specimens collected to the adult stage and make the species-level identification from the adults. This is a time-consuming process and often is not successful for a variety of reasons. Recent work has centered on the use of DNA for identifications of maggots [25, 26]. Although considerable work is yet to be done in characterizing the different species, improved techniques have significantly reduced both the time involved and the costs of such testing. In the relatively near future, this will become a widely employed technique for identification of early instars.

The other use of DNA material lies in the linking of suspects to victims based on DNA present in gut contents of parasitic insects. Repogle et al. [18] have demonstrated the potential for identification of DNA from gut contents and fecal material of the cab louse, *Pthirus pubis*. Additional studies have demonstrated the potential to determine hosts from gut contents of other insects such as mosquitoes. DiZinno et al. [7] have demonstrated the potential for recovery of mitochondrial DNA from beetle larvae recovered from human bone. Although the applications of these techniques are limited, in those cases where the application exists, this can be a valuable tool.

17.5.7 Abuse/Neglect of Children and the Elderly

Abuse/neglect of children and the elderly is one of the few instances in which techniques of medicolegal forensic entomology are applied to cases involving living

individuals. In the vast majority of instances, this involves a phenomenon known as myiasis. Myiasis is the feeding by maggots on living tissue or dead tissues associated with a wound. There are several subdivisions of myiasis and several routes for its evolutionary development have been hypothesized. In some instances, the species have evolved to the point where the larvae must feed on living tissues in order to complete their development to the adult stage, termed Obligatory Myiasis. This is encountered in a number of specie in the families *Calliphoridae*, *Sarcophagidae*, *Gasterophilidae*, *Oestridae*, and *Cuterebridae*. In some species, considerable damage is done to the host and death may result as a result of the infestation, as in the Screw worm fly *Cochliomyia hominivorax*. Others, such as the Human Bot fly, *Dermatobia hominins*, cause only localized discomfort and only limited damage to the host. Other species are found that feed on dead bodies or, occasionally, dead tissues associated with a wound on a live host. This is termed Facultative Myiasis and is what is most frequently encountered in legal situations. Because the larvae feed only on dead tissues, they cause little if any discomfort to the host and depart the wound when they have either completed development or there are no dead tissues remaining.

Prior to the introduction of antibiotics, "maggot therapy" was an accepted medical technique. It was first observed by Napolean's battlefield surgeon, who noted that individuals left on the field until maggots infested their wounds had a better chance of survival than those brought immediately into field hospitals. During the United States Civil War, the Confederate surgeon Zacharias was known to deliberately introduce maggots into wounds to clean the wound and prevent infections. The process became more refined and generally accepted until the introduction of antibiotics. As there have developed drug-resistant strains of bacteria, the technique has seen a resurgence [20].

In the forensic context, myiasis is most frequently associated with the facultative parasites in the families *Calliphoridae*, *Sarcophagidae*, and *Muscidae*. However, some use may be made of species demonstrating the obligatory state, such as *Dermatobia hominis (Cutrebridae*). If not fully appreciated, myiasis can be a significant point of confusion for the forensic entomologist, appearing to give an estimate of the PMI far longer than the actual period of time since death. In other instances, particularly in cases involving the living, an understanding of myiasis may prove to be a significant factor in resolving the case. In this chapter, several different situations involving myiasis will be demonstrated using case studies. Although myiasis is involved in both human and veterinary aspects of forensic investigations, in this treatment, only human involvement is presented.

In one case, specimens were submitted from the remains of a woman, 58 years of age, who had been in home care on the island of Oahu, Hawaii, in an extended family situation (two daughters, a son-in-law, and five grandchildren). The decedent had a history of stroke with right-side paralysis and only minimal contact with healthcare professionals. She was described as "difficult" and often remained in a wheelchair for extended periods of time, refusing to speak to family members for periods of several days. Specimens were collected at 09.30 on 10 July and submitted to the laboratory for analysis. Submitted specimens consisted of 3rd instar

larvae of *Phaenicia sericata* and egg masses of *C. megacephala*. The 3rd instar larvae of *P. sericata* indicated a mode developmental time of fifty hours, based on conditions inside the house. Based on the idea that the species showing the greatest period of residence on the body is indicative of the minimum period of time since death, this indicated an onset of insect activity at approximately 0800 on 8 July. By contrast, the eggs of *C. megacephala* hatched at 1400 on 10 July, indicating they had been deposited at approximately 0600 on 10 July. The family members stated that the decedent had last been seen alive at 0100 on 10 July.

Examination of the body during autopsy revealed the presence of a large necrotic area on the lower back, penetrating into the abdominal cavity. Maggots of *P. sericata* were restricted to this area, while the egg masses of *C. megacephala* were recovered only from the nasal cavities. The prosecutor in this case felt that this supported the account of the family as to the possible time of death. Although the presence of the 3rd instar larvae of *P. sericata* indicated an instance of neglect and a general lack of care for the decedent, charges were not filed by the prosecutor. Lacking the data concerning distribution of the maggots with respect to the wound and the involvement of *P. sericata* in myiasis in Hawaii, the estimated PMI would have been considerably longer than was actually the case. Too often, specimens are submitted by law enforcement agencies as a single collection from the remains, with no indication of location on the remains of infestations. If pre-existing infested wounds are not noted by those individuals making collections from the remains, the estimated period of time since death could be significantly in error.

Another case involving a still-living victim was described by Goff et al. [10]. In this instance, a 16-month-old child was discovered on the edge of Lake Wilson on the island of Oahu, Hawaii. The child was in a clear area surrounded by heavy vegetation. When found, she was suffering from dehydration and bruising and she had numerous insect bites. Initially, the period of exposure was estimated as being two days. Given the state of dehydration, a pediatrician suggested that the period was longer and that the child would most probably have died within the next 24 hours. The child was clothed in a sweatshirt, t-shirt, a pair of pants, and disposable diapers. On the front of the pants, from the waistband to a point below the knees, there were egg masses of a *Calliphoridae*. When the clothing was removed, numerous 1st instar larvae (measuring 3–4 mm total length) and fewer 2nd instar larvae (measuring 5 mm total length) of *C. megacephala* were discovered in the diapers and pants. Additional 1st and 2nd instar larvae of this same species were recovered from the vagina and rectum of the child and appeared to be feeding on tissues at those sites. Rearing data for this species from controlled studies conducted by Goff (unpublished data) at 26 and 28°C indicated the most mature larvae would have required 39 and 36 h, respectively, to reach the stage of development represented. Using ADH calculations without a base temperature to adjust for normal body temperature, it was estimated that it would have required 23.5 h to reach the most mature stage of development for the specimens collected from inside the diapers.

17.6 Educational Requirements and Certification

Obviously, forensic entomology is a highly specialized area of investigation, requiring a detailed knowledge of insect biology and taxonomy. With this in mind, those working in the field require specialized training. At present, there are no curricula available at universities in the United States offering degrees in forensic entomology. Rather, those interested in pursuing the field will be majoring in Entomology, Biology, or related fields, such as Forensic Sciences or Ecology. Although some workers in the past have begun working in forensic entomology with only a baccalaureate degree in one of these majors, the present trend is to view the Master of Science degree as the minimum level of education to adequately function in forensic entomology. The vast majority of workers in the United States currently hold an earned Doctorate in Entomology or Biology. During their graduate programs, the candidates will take extensive course work in taxonomy, ecology, and biology of insects. The thesis or dissertation topics, while filling all of the requirements of the granting institution, will have an emphasis on forensic aspects of the study, along with the more basis concerns.

Unlike many other areas of the forensic sciences, accreditation has only recently become available in Medicocriminal Forensic Entomology. The areas of Stored Product and Structural/Urban Forensic entomology fall under the purview of the Entomological Society of America through their Board Certified Entomologist Program, and accreditation has been available for many years. In 1996, the American Board of Forensic Entomology was created as an independent board. Certification is offered at two levels by this Board: Member and Diplomate. Member and Diplomate statuses are currently available to individuals filling the requirements from the United States and Canada. Individuals from other areas may become Associate Members, but no certification is associated with this level of membership.

Requirements for Member status include an earned M.S. degree in the field in Entomology, Biology, Ecology, or Zoology. The individuals must have a minimum of 5 years of professional experience following completion of their degree, with the last 3 years of the experience involving a substantial amount of work in medicocriminal forensic entomology. Evidence must be presented at the time of application of publication of relevant work in peer-reviewed journals, presentations at meetings, and submission of case studies. Requirements for Diplomate status are the same as for member, but the applicant must possess an earned Doctorate in one of the listed disciplines. Applicants for both Member and Diplomate status are required to complete a written and practical examination, passing with a minimum score of 80%.

There is no accreditation available for laboratories in the area of Forensic Entomology. Currently, forensic entomologists are either housed in academic institutions where a significant portion of their research is involved with Forensic Entomology or employed in crime laboratories with other duties in addition to forensic entomology.

17.7 Appendix: Protocols for Collection of Entomological Specimens

The insects and other arthropods associated with a decomposing body are a potentially powerful tool in a homicide investigation. Their ultimate utility, however, depends on their being properly collected and preserved for analysis by an entomologist. The ideal situation would entail the collection and shipment of a single specimen. This would be collecting and shipping an entomologist to the scene. The entomologist would then examine the body and make all of the necessary collections. Whenever possible, local law enforcement should consider establishing a working relationship with an entomologist from a local college or university. This individual may not be familiar with the techniques of forensic entomology, but will be familiar with the local arthropod fauna and correct collection and preservation techniques. This will assure that the specimens are properly handled. Because I realize this may not always be possible, this protocol is designed to assist the non-entomologist in proper collection, preservation, and shipment of these specimens.

17.8 Equipment Needed

1. Surgical gloves
2. Forceps
3. Glass vials
4. Trowel
5. Insect net
6. Small artist's brush
7. Paper bags
8. Plastic bags and paper bags
9. Ice cream cartons
10. KAA or other insect fixative
11. Vermiculite or sand
12. Ethyl alcohol (70–80%) or isopropyl alcohol
13. Can of cat food

17.9 Collection Procedures

1. Flying insects

Prior to collecting insects and other arthropods, it is advisable to have personnel move away from the body for a period of ten to fifteen minutes. While many of the flies and beetles that may be present on the body are quite determined and will

remain during scene processing, there are other significant species that will leave the body when it is disturbed. If the body is left alone, these insects may return. It is important to make collections of flying insects as soon as possible to assure that a representative sample is collected. These insects can be collected using the insect net. An insect net is relatively easy to use, but I would suggest some practice before using a net at the scene. This will help to preserve some of your dignity. Remember, you have just produced an insect net and announced that you are going to collect flies. Reactions to this are varied under the best of circumstances. Samples should be taken from the body and also any adjacent bushes that may serve as refuges for insects driven from the body. Insects collected in this manner will usually be adults and hard bodied. These should be preserved in glass vials using the 70–80% ethyl alcohol or the isopropyl alcohol. If the isopropyl alcohol is used, it should be cut 1:1 with water. Otherwise the insects will become hardened and difficult for the entomologist to identify. Formalin should not be used to preserve insects unless there is no alternative available. If preserved in formalin, insects should be transferred to ethyl alcohol as soon as possible. Samples from different areas (over the body, in bushes, etc.) should be kept separate and labeled as to origin, collector, how collected (net, picked up by hand, etc.), and time of collection. This information will be very important to the entomologist.

2. Crawling insects

Insects will be crawling on or in a number of different places on the body. Collections made from each part of the body should be kept separate and labeled as to their origin. These insects can be collected using the forceps or by hand. You should wear the surgical gloves at all times while sampling from the body. The arthropods you collect in this manner will be of two general kinds: (1) hard-bodied adults, such as beetles, and (2) immature and soft-bodied insects. The hard-bodied insects can be treated in the same manner as the insects collected using the insect net. The soft-bodied insects and immature insects require some special treatment to ensure that they will be suitable for analysis by the entomologist.

Immature insects are frequently difficult to identify, whereas the adults can be easily identified by an entomologist. Even closely related species may have different patterns and rates of development. Correct identification of the species is essential to the entomological investigation. For this reason, it is desirable to keep some of the immatures alive to be reared to the adult stage. I generally suggest splitting the collections into two parts. The first will be killed to stop the biological clock that will be analyzed by the entomologist and the other kept alive to confirm the species identifications.

The immature insects, most frequently maggots, to be preserved for later analysis should be killed and preserved in vials of a KAA solution for a period of five to ten minutes, depending on the size of the maggots, and then transferred to 70% ethyl alcohol or isopropyl alcohol cut 1:1 with water for storage. The KAA solution is designed to breakdown the waterproofing on the insect's cuticle or outer body surface. If this is not done, the alcohol will not penetrate the body and the insect will become blackened and rot. The KAA solution consists of 1 part glacial acetic

acid, 1 part refined kerosene, and 30 parts 95% ethyl alcohol. There are other insect preservative solutions available, but this is the easiest to use. If KAA is not available, the insect can be fixed using hot water. In this case, the maggots are placed into hot water of approximately 76.7°C (170°F) for a period of 2–3 min and then transferred to 70% ethyl alcohol for storage. Water near this temperature can be obtained from most fast food restaurants by ordering tea or decaffeinated coffee.

The rearing of immature insects to the adult stage requires a set of controlled conditions and an adequate food source. Rearings are frequently essential for accurate determinations of the species involved in the decomposition process, because the immatures of many species are virtually indistinguishable.

Immatures collected from the scene to be reared to the adult stage should be placed into the ice cream containers. I generally suggest a 1/2 pint container (0.95 L). This container should be filled 1/4 to 1/2 with the vermiculite or other inert material. In selecting your material, be careful not to use anything that has an insecticide added. Moist soil can be used for this purpose if other materials are not available. A food source is advisable and a small amount of the canned cat food or beef liver will serve this purpose. Do not place specimens to be reared or containers with these specimens into plastic bags or tightly sealed vials. They will not survive these conditions, particularly in warmer weather. These specimens should be transported to the entomologist as quickly as possible.

An entomologist will normally have facilities available for rearing of immatures and those specimens collected from a crime scene should be transported to the entomologist as quickly as possible. Generally, normal mail should not be used for transport of live specimens from a crime scene. Package all containers in well-padded shipping containers to avoid breakage. Vials should be individually wrapped and placed in a box with at least two inches of Styrofoam chips surrounding all sides, top, and bottom. Package soil samples and other living specimens in such a manner as to ensure they will remain as cool as possible and in well-ventilated containers. DO NOT ship living materials in plastic bags under any circumstances. Soil samples can be shipped and stored in paper bags that are folded over at the top and stapled shut.

If the specimens cannot be shipped to an entomologist immediately, the larvae can be reared to the adult stage in the ice cream containers if provided with a sufficient quantity of food. For this purpose, I suggest placing the larvae and food material into a small watch glass placed inside the container. The top of the container should be covered with a fine gauze or organdy material held in place with a rubber band around the outside of the container to allow for air and easy observation of the larvae. The larvae should be checked daily and additional food added as needed. There should be as little disturbance of the container as possible. The larvae will complete their development on the food source and then migrate away from the food source, downward into the vermiculite to pupate. Adults will emerge from the puparia and crawl to the surface. The conditions under which the larvae are reared should be as close as possible to those of the actual crime scene to allow for accurate determination of the times required for development. Temperatures (daily maximum/minimum) should be recorded for the area in which the larvae are reared. Emerging adults sold be provided with food in the form of a cotton ball soaked with water and sugar. They should be

allowed to feed for a 24 hour period, then killed and either preserved in 70% ethyl alcohol or dried and pinned. This allows for the exoskeleton to dry and accurate determinations can then be made by the entomologist.

17.10 Labeling

Each lot of specimens collected should be well labeled to ensure proper interpretations of the evidence by the entomologist. Each container should be labeled separately and include the following data:

1. Date collected
2. Time collected
3. Location of body – be as specific as possible
4. Type of habitat – inside house, in bushes, on side of hill, etc.
5. Location on body of specimens collected – DO NOT combine collections from different parts of body
6. Name, address, and telephone number of collector

17.11 Additional Information

A description of the locality is essential to the proper interpretation of the insect evidence. Insects are frequently quite specific as to the type of habitats in which they will be found and their activities may vary from one habitat type to the next. General photographs of the scene are invaluable to the entomologist. While videotapes are of some use, the 35-mm still photograph is currently of the greatest use to an entomologist in analyses. Your written description of the locality should include the following:

1. The geographic location – city, county, street address if applicable, etc.
2. General type of habitat – desert, forest, pasture, inside apartment, dump, etc.
3. Terrain – rocky area, hill side, flat area, etc.
4. Type of vegetation present – samples, if feasible, should be sent to a botanist
5. Soil type – sand, gravel, mud, artificial (cement, blacktop, etc.)

A description of the corpse along with a detailed photographic record. The description of the corpse must include the following:

1. Sex, height, and weight
2. Presence or absence of clothing and description of clothing
3. Orientation of corpse – sitting, lying face down, lying on back, etc.
4. Any attempt to conceal the corpse – wrapping, covered by vegetation, etc.
5. Physical damage – lacerations, scrapes – and location of damage
6. Cause of death
7. State of decomposition
8. Insect fauna observed. Include close-up photographs if possible

Complete climatic data from the NOAA weather station closest to the crime scene and data from any other weather stations (small airports, agricultural experiment stations, etc.) in the vicinity that may be closer to the scene. These should include rainfall and temperature as a minimum. Hourly temperature data are desirable. Insect development is affected by many different factors, but weather factors, particularly temperature, are among the most significant. These govern adult activity, including egg laying, and immature development.

Finally, anything unusual about the scene of general area should be noted. If possible, make a photographic record of any unusual aspects of the scene. A complete photographic record of the crime scene is invaluable to the entomologist, often showing factors that may be of significance, but not obvious to the non-entomologist processing the scene.

As I stated at the beginning, entomological evidence has the potential to be a powerful tool in a criminal investigation. It may well not be present in all cases. When it is present, it will be reliable only if the evidence is properly collected and documented prior to being turned over to the entomologist for analysis. As with all other types of physical evidence, a proper chain of custody must be maintained during the processing.

17.12 Questions

1. What attributes of insects allow for their use in the estimation of a period of time since death and what is actually estimated?
2. What are the subdivisions of what is commonly termed "Forensic Entomology?"
3. When collecting immature insect specimens from a body, the collections are split into two lots. Why is this done and what is done with each lot?
4. What are the two aspects of Entomotoxicology that must be considered in a death investigation dealing with remains during the later stages of decomposition?
5. How can insects assist in the evaluation and/or detection of wounds on a body during early stages of decomposition?
6. What is the Accumulated Degree Day concept and how is this used in forensic entomology?
7. How is DNA currently employed in medicocriminal forensic entomology?
8. How can insect activity assist in cases of abuse and neglect of children and the elderly?
9. What body provides accreditation for medicocriminal forensic entomology in the United States and Canada?
10. What would be the minimum level of education required for a forensic entomologist?
11. In what way can insects assist in determining if a body has been moved following death?

17.13 About the Author

M. Lee Goff received his B.S. degree in Zoology from the University of Hawaii at Manoa in 1966, his M.S. in Biology from California State University, Long Beach in 197, and his PhD in Entomology from University of Hawaii at Manoa in 1977. The early part of Dr. Goff's professional career was spent working at the B.P. Bishop Museum on the biology and taxonomy of the larval Trombiculidae and Leeuwenhoekiidae as well as the role of avian malaria in the decline of endemic Hawaiian land birds. He began working in Forensic Entomology in 1983, while still at the B.P. Bishop Museum in Honolulu. He left that position and moved to the Department of Entomology in the University of Hawaii at Manoa in late 1983. While there, he was a professor of Entomology and Chair of the Entomology Graduate Program. He left the University of Hawaii in August 2001 for Chaminade University of Honolulu, where he is currently Director of the Forensic Sciences Program. He has published more than 225 papers in scientific journals dealing with Acarology, Medical Entomology, and Forensic Entomology and authored the popular book "A Fly for the Prosecution." He has conducted numerous workshops dealing with applications of entomology to forensic problems in different venues around the world, including presentations for the FBI Academy in Quantico, Virginia. He has participated in numerous death investigations and provided expert testimony. Dr. Goff is a Fellow of the American Academy of Forensic Sciences, Affiliate Member of the National Association of Medical Examiners, American Association of Clinical Laboratory Directors, and a Diplomate of the American Board of Forensic Entomology. In addition, Dr. Goff has served as a consultant for numerous television shows, including the popular CSI Las Vegas, CSI Miami, and Bones.

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